

ETHANOLIC EXTRACT OF POLISH PROPOLIS (EEP) IN REACTION WITH FREE RADICALS

Czuba Z.P., Seget S., Krol W.

School of Medicine with the Division of Dentistry in Zabrze, Medical University of Silesia in Katowice, Chair and Department of Microbiology and Immunology, Poland

Propolis (bee glue) is a resinous product found in bee hives where it is used by honeybees as cement and to seal cracks or open spaces. Honeybees collect propolis from tree buds and other botanical sources. The chemical composition of propolis varies considerably from region to region along with vegetation and from season to season. Among other properties propolis shows antioxidant activity. It is mainly depended on concentration of many phenolic compounds, flavonoids and their derivatives. Complex and not fixed chemical composition can influence the antioxidant property. In our study an ethanolic extract of Polish propolis (EEP) was used.

Antioxidant activity was measured applying three methods:

1. DPPH radical scavenging activity

To 0.04 ml test sample was added 0.120 ml methanol. Then the sample was mixed with 0.04 ml DPPH in methanol about ($A_{524\text{nm}} = 0.900 \pm 0.020$). After 15 min the optical density of the sample was measured at 524 nm. Next, we set the curve for Trolox and EEP.

2. ABTS radical scavenging activity.

ABTS was dissolved in distilled water to a 7 mM concentration. ABTS radical cation ($\text{ABTS}^{\bullet+}$) was produced by reacting ABTS stock solution with 2.45 mM potassium persulfate (final concentration) and allowing the mixture to stand in the dark at room temperature for 16 h before use. Then a solution of 0.180 ml of dilute $\text{ABTS}^{\bullet+}$ ($A_{734\text{nm}} = 0.700 \pm 0.020$) was added 0.2 ml test sample. Then the sample was incubated 15 min. The optical density of the sample was measured at 734 nm. Next, we set the curve for Trolox and EEP.

3. Ferric reducing-antioxidant power (FRAP) assay

To 0.180 ml of the working compound (0.3 M acetate buffer pH 3.6, 10 mM 2,4,6-Tris (2-pyridyl) -s-triazine TPTZ 40 mmol/l HCl; 20 mM FeCl_3 , in ratio of 10:1:1) was added 0.02 ml test sample. After 15 min the optical density of the samples were measured at 593 nm and set the curves.

The obtained result:

EEP: $\text{ID}_{50 \text{ DPPH}} = 13.9 \mu\text{g/ml}$; $\text{ID}_{50 \text{ ABTS}} = 8.4 \mu\text{g/ml}$

Trolox: $\text{ID}_{50 \text{ DPPH}} = 46.0 \mu\text{g/ml}$; $\text{ID}_{50 \text{ ABTS}} = 141.4 \mu\text{g/ml}$.

The used methods could be helpful in standardization of propolis extracts.